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Corrections

The authors regret making an unfortunate error in their article on "Filter-Press Cake as a Fertilizer," THE JOURNAL OF AGRICULTURE **39** (4) 198–213, 1955. On p. 206 under the subhead "Pineapples," the work credited to Hernández-Medina, *et. al.* (1), working on a Bayamón silty clay at Manatí, was actually performed on a Bayamón silty clay at Palo Blanco, Arecibo, as stated in line 9 under the aforementioned subhead. George Samuels and Pablo Landrau, Jr.

In the Literature Cited section of the paper by J. Adsuar and L. López Matos, entitled "Reaction of some Nicotiana Species to the Pepper- and Common Tobacco-Mosaic Viruses," JOURNAL OF AGRICULTURE **39** (3) 171, the first name in reference 2 is given as Pérez Otero, E., instead of correctly as Pérez, J. Enrique. In reference 3 to the Research Note on "Cross-Protection Tests Confirm the Presence of Etch Virus on Tobacco in Puerto Rico," by Adsuar and Pérez, THE JOURNAL **40** (1) 83, as well as at the end of the article on p. 84, the name reads J. E. Pérez in lieu of J. Enrique Pérez. Only one person, not three persons, is involved.

J. Enrique Pérez

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J. Enrique Pérez

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Identification of Sugars Present in Fruit of the Acerola (*Malpighia punicifolia* L.) by Paper Chromatography

R. Santini, Jr., and Alice S. Huyke[1]

INTRODUCTION

Nothing is found in the literature regarding the sugars present in the fruit of the acerola. The identification of these sugars is of great importance for the study of the reactions which take place when the juice is flash-pasteurized and canned.

Paper chromatography is an important tool in analytical chemistry and has been used by many investigators to identify the sugars present in various plant materials. The qualitative paper-chromatography method of William and Bevenue (1)[2] was employed for the work on the acerola.

Before applying it as described below, some preliminary work had been done at this Station to identify the sugars present in the acerola. The work was initiated using the method of Moore and Link (2), but no definite results were obtained.

A phenyl osazone derivate was prepared using the sugar extract of the fruit. This osazone was identical with the one obtained from dextrose, levulose, or mannose. The Seliwanoff (3) and cobaltous chloride (4) tests were positive for levulose. However, the quantitative determination of levulose using the Jackson-Mathews modification of the Nyns selective method (5) indicated that it comprises only about 50 percent of the sugar present in the fruit.

Some work was also done on the determination of reducing and total sugars in acerola juice using the Lane-Eynon method (6). The statistical analysis of the results revealed that, if sucrose is present in the juice, it must be in quantities so small that it cannot be determined by this method.

PROCEDURE

Three hundred grams of acerolas were chopped and quickly added to sufficient 95-percent ethyl alcohol to give, with the water naturally present

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[2] Numbers in parentheses refer to Literature Cited, p. 89.

in the fruit, an approximate alcohol concentration of 80 percent. One to two grams of $CaCO_3$ were added and the mixture was heated for 1 hour on a steam bath. The supernatant liquid was filtered and the residue was covered with 80-percent ethyl alcohol, reheated for 30 minutes on the steam bath, and filtered. The filtrates were then combined.

An aliquot containing 4 gm. of sugars was evaporated on a steam bath to remove the alcohol. The concentrate was filtered through a mat of Celite analytical filter-aid and the filtrate was made to 100 ml. with distilled water.

To remove the substances that interfered with the identification of sugars by paper chromatography, the solution was passed first through a column containing the cation-exchanger Amberlite IR-120 and then through a column containing the anion-exchanger Amberlite IR-4B. The pH of the eluate was 6.8. The deionized solution was then ready to be chromatographed.

A penciled line was drawn parallel to, and about 8 cm. from one end of the paper (Whatman No. 1, 18½ inches x 22 inches). Division marks about 2 cm. apart were made on this line. The other end of the paper was trimmed with pinking shears to give a saw-toothed effect.

Three-percent solutions of galactose, dextrose, mannose, sucrose, sorbose, levulose, arabinose, ribose, fucose, raffinose, xylose, lyxose, maltose, and lactose were prepared to be used as standards.

Applications of known sugar solutions (5 λ) and acerola extracts (10 λ) were made on the division marks (2 cm. apart) on the paper.

Three chromatograms were irrigated with a mixture of 8 parts of ethyl acetate, 2 parts of pyridine, and 1 part of water. After 16 hours the chromatograms were ready to be dried in the hood and sprayed.

The spray reagents used were resorcinol, aniline, and 3,5 dinitro salicylic acid. Resorcinol is specific for the detection of ketoses and polysaccharides which contain ketose sugars. Aniline gives a purple color with pentose and a brown color for the other sugars, and 3,5 dinitro salicylic acid is used for the detection of aldoses, pentoses, and some disaccharides, but does not detect levulose-containing sugars such as raffinose and sucrose.

RESULTS

The spots obtained with the acerola extract corresponded to dextrose, levulose, and sucrose. However, since levulose and sorbose have such very close Rf values in this solvent, the work was repeated using another solvent mixture consisting of 4 parts of butanol, 1 part of acetic acid, and 5 parts of water.

Two phases were formed. After the mixture had been shaken and al-

lowed to stand several hours, the H_2O-rich phase was added to the bottom of the chromatographic cabinet and the alcohol-rich phase was added to the troughs. The spots obtained with this solvent mixture and the same spray reagents mentioned above confirmed the presence of levulose, dextrose, and sucrose in the acerola.

SUMMARY

By means of paper chromatography it proved possible to identify the sugars in the fruit of the acerola. Dextrose, levulose, and sucrose were found to be present.

RESUMEN

Por medio de la cromatografia de papel fué posible identificar los azúcares presentes en la acerola. Dextrosa, levulosa, y sacarosa fueron los azúcares que se encontraron presentes.

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Occurrence of Pectin Methylesterase in Guavas

Irma G. Rieckehoff and A. Ríos[1]

INTRODUCTION

Pectin methylesterase has been found to be present in the fruits, leaves, stems, and roots of various plants. Citrus fruits and tomatoes (1)[2] are among the fruits mentioned in the literature as showing high activities of the enzyme.

The formation of gel-like particles in guava nectars has been observed for some time in this laboratory. When cans of this product are opened, the gel-like particles can be seen suspended in the liquid. These solid particles, which cannot be disintegrated by stirring the nectar well, detract from the appearance of the product.

Rouse (2) reported that gelatinization of citrus concentrates is caused by the action of pectin methylesterase, which partly demethylates the pectin present to form low methoxyl pectin that produces gels in the presence of polyvalent cations. Wenzell, Moore, Rouse, and Atkins (3) subsequently showed that the pectin content, enzyme activity, and the concentration of polyvalent cations were the principal factors responsible for gel formation in citrus concentrates.

The presence of methylesterase in guavas has not been reported before. Since guava nectars of the Dominica Roja variety contain approximately 0.003 to 0.004 percent of Ca (4) and about 0.3 percent of pectin (as pectic acid), the pH varies from 3.3 to 3.7, and the soluble solids are adjusted to 15° Brix, it was suspected that the formation of gels in the nectars might be due to a reaction similar to that already reported as taking place in citrus concentrates. It was decided to investigate the occurrence of pectin methylesterase in guavas to obtain more information about the possible change taking place in the nectars.

EXPERIMENTAL PROCEDURE

Whole, ripe, firm guavas of the Dominica variety (both sweet and sour types) were separated by hand into three parts: Peel, shells and inner pulp, and seeds. Each part was homogenized separately in a Waring Blendor and stirred for about 3 minutes with 3 parts of a 0.35-M solution of NaCl. The pH of the mixtures was adjusted to 8 with dilute NaOH and the dispersion left overnight at 50°F. The next day they were filtered and 20 ml. of the filtrate were used for the assay. The method used was essentially that pro-

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[2] Numbers in parentheses refer to Literature Cited, p. 92.

posed by Kertesz (6). Each 20 ml. of the extracts corresponded to 5 gm. of the original wet tissue and to 0.75 gm. of dry tissue. Fifty milliliters of a 1-percent solution of Exchange citrus pectin were added. The reaction medium was brought in each case to pH 6.2. The pH was kept constant at this value by the addition of 0.05 N NaOH with a microburette at 10-minute intervals for 30 minutes, at the end of which time the mixture was rapidly titrated to pH 7.5. The temperature of the reaction medium was 26°C.

Controls were run by heating the extracts for 3 minutes at 190°F. and treating them exactly like the samples. The differences between the volumes of 0.05 N NaOH needed to titrate the samples and the controls to pH 7.5 were taken as the volumes of 0.05 N NaOH which were used by the samples as a result of enzyme activity. Pectin methylesterase units are expressed as milligrams of OCH_3 per gram of original wet tissue per 30-minute reaction time. The results of the assays are presented in table 1.

TABLE 1.—*Methylesterase content of different parts of guavas of the Dominica Roja variety*

Part tested	NaOH consumed	P. E. units[1] per gram of wet tissue	P. E. units[1] per gram of dry tissue
	Meq.	*Mg.*	*Mg.*
Peel	0.080	0.49	3.27
Shell	.010	.06	.40
Pulp and seeds	.026	.16	1.07

[1] Expressed as milligrams of OCH_3 per gram of wet tissue per 30 minutes.

DISCUSSION

The results in table 1 show a noticeable methylesterase activity in guavas. However, this activity is very much less than that of tomatoes, citrus flavedo, etc. The optimum conditions for extraction of the enzyme from the tissues of the guava are not known. The temperature at which the demethylations were carried out was much less than the optimum for enzymes separated from other sources. A complete study of the extraction procedure and the optimum conditions for action of the methylesterase of guavas will soon be undertaken in this Laboratory.

No activity could be found in an extract from the leaves of the guava tree, nor could any be found in the clear filtered juice of the fruit. The highest enzyme activity is apparently located in the peel of the fruit, while the shell shows the least.

The conditions present during processing of guava nectars appear to be favorable to the demethylation of the pectin and subsequent formation of low-methoxyl gels. It takes at least 1 hour in good commercial practice for the guava nectar to be canned, from the time the guavas are ground until

the moment the nectars are flash-pasteurized. Although enzyme activity seems to be low, the other factors necessary for the formation of low-methoxyl gels are present in the fruit dispersions. Pasteurization procedures used for guava nectars appear to be adequate since no enzyme activity could be detected in the pasteurized nectars.

SUMMARY

It has been possible to detect the presence of pectin methylesterase in guavas. It is suggested that partial demethoxylation of the pectin in the nectars and subsequent formation of insoluble pectinates with bivalent cations, may be responsible for the formation of insoluble gel-like particles present in some canned guava nectars.

RESUMEN

Se ha podido demostrar la presencia de la metilesterasa de pectina en la guayaba. Se sugiere que la demetoxilación parcial de la pectina presente en los néctares de guayaba y la subsiguiente formación de pectinatos de iones bivalentes pueden ser responsables de la presencia de los coágulos insolubles que se encuentran en algunos néctares de guayaba enlatados.

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A New Instrument for Measuring the Breaking Strength of Jellies

Irma G. Rieckehoff[1]

INTRODUCTION

Several instruments have been described in the literature for measuring the firmness of jelly. (1, 2)[2] Some of these measure firmness without exceeding the elastic limit of the jellies; others measure the breaking strength of the jellies (3). In the past there has been considerable disagreement as to the characteristics that should be measured in test jellies. Recently it has been advocated that both the rigidity and breaking strengths be measured when grading pectins (4). While both the Exchange Ridgelimeter developed at the California Fruit Growers Exchange and the Rigidometer developed at the Western Utilization Research Branch, USDA, are fairly satisfactory for measuring rigidity, the Tarr and Baker Tester, which is almost exclusively used for determining breaking strength, is not altogether satisfactory. The difficulties presented by the inability to apply a uniform pressure and the effect of the skin formed on top of the jellies have not been obviated.

The breaking strength of jellies is a more important characteristic from the preserver's point of view than the rigidity. The manufacturer of jellies is more interested in knowing how well his jellies will stand rough handling in shipping to distant points and how well a portion of jelly will spread over a piece of toast, cake, etc. For control work, the simplicity and reproducibility of a method are very important assets.

A torsional viscometer with a special attachment which lowers the rotating spindle into the material at a uniform rate has been found very convenient for measuring the breaking strength of jellies. The drag produced upon a spindle rotating at a definite constant speed while immersed in the jelly is recorded. At the same time, the spindle rotates as it is lowered into the body of the jelly. The torque required to turn the spindle as it constantly encounters new material is indicated on the dial of the viscometer. Since jellies are non-Newtonian in rheological properties the instrument will not give a true viscosity, but really a viscosity at a rate of shear which depends on such factors as the size of the spindle, the velocity of the rotating shaft, and the temperature. The instrument can be used at four different speeds and there are six different spindles for various ranges of viscosities of the

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[2] Numbers in parentheses refer to Literature Cited, p. 100.

materials examined.[3] Figure 1 shows the instrument assembled for use, and figure 2 the set of spindles which come with the instrument.

FIG. 1.—Torsional viscometer of the type used in this study.

EXPERIMENTAL PROCEDURES

The first step in the testing of the instrument consisted in selecting the most convenient speed and spindle to be used in the measurement of jelly strength. A guava jelly pronounced of excellent firmness by a panel of ex-

[3] The instrument used in these studies is manufactured by Brookfield Engineering Laboratories, Inc., Stoughton, Mass. It is calibrated in two scales: 100-unit and 500-unit.

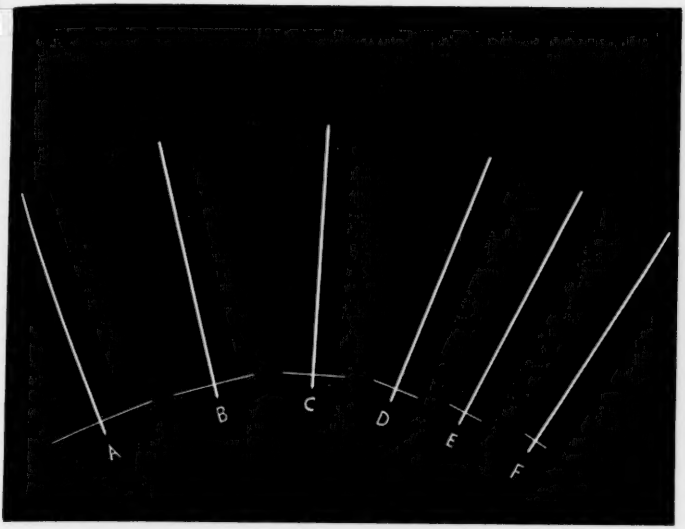

FIG. 2.—Set of spindles used: A, Spindle A; B, Spindle B; C, spindle C; D, spindle D; E, spindle E; F, spindle F.

TABLE 1.—*Readings of the viscometer and approximate viscosities in centipoises*[1]

Spindle	Velocity	Viscometer readings	Average reading	Average
	R.p.m.			*Centipoises*
C	1	45.0, 37.5, 38.0	40.2	804,000
C	2	60.0, 59.0, 52.0, 52.0, 43.0, 43.0	51.5	490,000
C	5	70.0, 65.0, 70.0, 65.0, 65.0, 67.0, 60.0 60.0	65.3	261,200
C	10	—	—	—
D	1	18.0, 15.0, 18.0	17.0	680,000
D	2	25.0, 23.0 24.0 25.0, 25.0, 25.0	24.5	490,000
D	5	40.0, 40.0, 38.0, 38.0, 35.0, 35.0, 35.0 37.0	37.3	298,400
D	10	—	—	—
E	1	12.0, 10.0, 10.0	10.7	1,070,000
E	2	15.0, 12.0, 12.0, 12.0, 12.0, 12.0	12.5	625,000
E	5	17.0, 17.0, 18.0, 18.0, 15.0, 15.0, 15.0, 16.0	16.4	328,000
E	10	—	—	—
F	1	3.5, 3.0, 3.0	3.3	660,000
F	2	7.0, 5.5, 7.0, 5.0, 6.0, 5.0	5.9	590,000
F	5	9.0, 7.0, 9.0, 9.0, 7.0, 8.0, 9.0, 8.0	8.3	332,000
F	10	—	—	—

[1] Measurements were made at approximately 28°C. and readings are on the 100-unit scale.

aminers was selected to be tested using all four velocities and six spindles. Spindles A and B were eliminated by inspection because the crosspieces

TABLE 2.—*Reproducibility of the method of measuring jelly strength using spindle C, velocity 1 r.p.m., 28°C.*

Container No.	Viscometer readings	Average
1	42.0, 28.0, 31.0	33.9
2	36.0, 29.0, 32.0	32.3
3	38.5, 29.0, 30.0	32.5
4	37.5, 30.0, 32.5	33.3
5	43.0, 29.0, 30.0	34.0
6	35.0, 33.0, 26.5	31.5
Mean		32.92
Standard deviation		.98
Standard error		.40
Coefficient of variation		2.98 percent

TABLE 3.—*Readings of the viscometer when used on jellies in different types of containers*

Type of container	Viscometer readings	Average
6-oz. Hazel Atlas jars	1: 35.0, 27.5, 31.2	31.2
	2: 24.0, 25.0, 35.0	28.0
9-oz. screw-cap jars	1: 38.0, 27.0, 25.0	30.0
	2: 35.5, 27.5, 27.0	30.0
6-oz. tumblers with pressure caps	1: 28.0, 34.0, 40.0	34.0
	2: 24.0, 35.0, 31.0	30.0
12-oz. tumblers with pressure caps	1: 38.0, 27.5, 25.0	30.2
	2: 37.0, 28.0, 27.0	30.7
12-oz. tall jars with aluminum screw caps	1: 36.0, 28.0, 24.0	29.3
	2: 38.0, 28.0, 28.0	31.3
Mean		30.47
Standard deviation		1.56
Standard error		.49
Coefficient of variation		5.09 percent

were considered too long (48 mm. and 36 mm. respectively) to be used in small jelly jars. Spindles C, D, E, and F were used with velocities 1, 2, 5, and 10 revolutions per minute. The data obtained are presented in table 1.

Approximate viscosity values are given in centipoises. These values were calculated from the calibration values given in a table supplied with the instrument.

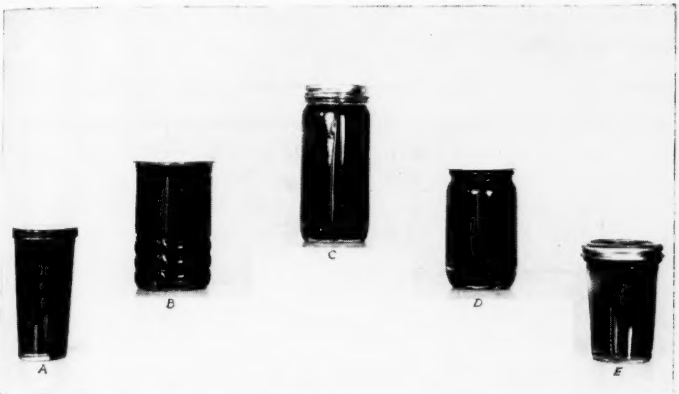

FIG. 3.—Types of jelly jars used in this work: A, 6-oz. tumbler with pressure cap; B, 12-oz. tumbler with pressure cap; C, 12-oz. tall jar with aluminum screw cap; D, 9-oz. screw-cap jar; E, 6-oz. Hazel Atlas jar.

TABLE 4.—*Comparison between values of jelly strength obtained by the torsional viscometer and the Delaware jelly strength tester*

Sample No.	Delaware instrument	Brookfield instrument
1	17.0	7.3
2	20.0	10.8
3	25.0	15.0
4	38.0	23.5
5	40.0	25.0
6	49.0	29.0
7	50.0	28.5
8	53.0	35.0
9	75.0	45.0

The reproducibility of the selected method of measuring jelly strength was tested by taking readings in six different jars of the same type containing jelly from a single batch. Results are presented in table 2.

The effect of size and shape of the containers, ranging from 6-ounce to 12-ounce jars was studied by pouring jelly of one batch into the following containers: 6-ounce Hazel Atlas jars, 6-ounce tumblers with pressure caps, 9-ounce jars with screw caps, 12-ounce tall jars with screw aluminum caps,

and 12-ounce tumblers with pressure caps. The data are given in table 3. Figure 3 shows the type of jars used for the above work.

The jelly strength of a series of jellies was determined both by the torsional viscometer and Tarr and Baker's Delaware jelly strength tester. Table 4 presents values obtained with both instruments. Figure 4 shows the relation between them.

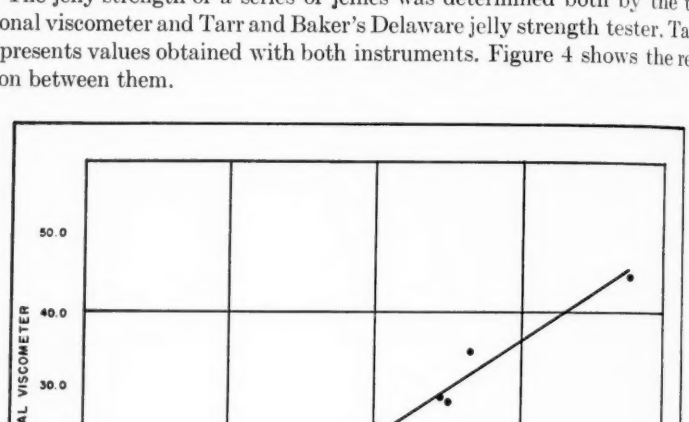

FIG. 4.—Relation between values of jelly strength measured with the viscometer and with the Tarr and Baker Delaware Jelly Strength Tester.

RESULTS AND DISCUSSION

From the data presented in table 1 it is evident that, since the jelly tested was of optimum firmness, the values obtained using spindle D at 1 and 2 revolutions per minute, spindle E at 1, 2, and 5 revolutions per minute, and spindle F at 1, 2, and 5 revolutions per minute were not convenient to use. It is desirable that a jelly of good firmness should read around 40 to 60 units so that the more tender and the tougher jellies will give satisfactory readings. Values for 10 revolutions per minute were not included because they were very erratic. It is not possible for the indicator needle to come to equilibrium at high speeds.

Good values were obtained using spindle C at 1, 2, and 5 revolutions per minute and spindle D at 5 revolutions per minute. Spindle C was chosen

over spindle D because it gave slightly higher values. The lowest speed was selected since the literature accompanying the instrument said that low speeds were preferable for such materials as jellies, because the pitch of the helical path described by the crosspiece as it is lowered into the jelly is greatest at the lowest speed.

The reproducibility of the method of measurement using spindle C at 1 revolution per minute is shown in table 2. The values are expressed as instrument readings rather than in centipoises because the calibration of the spindle ranges in centipoises is approximate. The first reading is usually the highest and quite different from the others taken from the same jar. This discrepancy in the first reading may be due, in part, to inability of the instrument to reach equilibrium before the first reading is taken and, in part, to a skin effect which is noticeable only at the top of the jelly. After the instrument reaches equilibrium the readings are fairly uniform. Measurements should be made in jars which allow at least three readings to be taken so as to minimize the influence of the first reading. The agreement between averages of three readings per jar is excellent. The measurement of jelly strength should always be determined by using duplicate jars and making at least three readings per jar.

The effect of size and shape of container is presented in table 3. It can be seen that capacity of containers from 6 to 12 ounces does not appreciably affect the values for jelly strength as measured with the viscometer. Although the variation is greater than when measurements are made in the same type of jar, it is still acceptable. When the diameter of the jars is not uniform, but diminishes toward the bottom, as in the case of both 6-ounce containers, the last readings usually increase. It is preferable to use containers of uniform diameter so as to get more uniform values.

The data in table 4 and figure 4 show the relation between the values obtained using the Delaware tester and the viscometer. Values around 50 to 60 units on the Delaware tester correspond to values of about 30 to 40 units on the viscometer. Tarr and Baker considered strengths of 50 to 60 cms. of water excellent as determined by their instrument. It has been found that jellies of optimum firmness give values of about 40 units in the type of viscometer used in our study.

The described method of measurement has a number of advantages over other methods of measuring the breaking strength of jellies. The skin effect is not important since readings are taken in at least three levels of the column of jelly. The rate of applying the shear is uniform when the same velocity is used throughout the measurements. The radius of the helical path described by the crosspiece as it is lowered into the column of jelly is constant when a definite spindle is used. It is not necessary to remove the jellies from their containers and any inexperienced operator can make

the measurements in any control laboratory. The reproducibility of the values is higher than that of the values obtained by the use of the Delaware tester.

SUMMARY

1. A torsional viscometer with a special device to lower the spindle of the instrument as it rotates has been used to determine the breaking strength of jellies and has been found excellent as to reproducibility of values obtained and simplicity of operation.

2. Values obtained by use of the tested viscometer are compared with corresponding ones obtained by the use of Tarr and Baker's Delaware jelly strength tester.

3. A torsional viscometer with a special attachment as described in this work is strongly recommended for control laboratories in the preserve industry. It is suggested that it be adopted for measuring the breaking strength of test jellies made to determine the jelly grade of pectins. A calibration curve can be prepared using a standard pectin and grades of tested pectins can be determined from such a curve.

RESUMEN

1. Un viscómetro de torsión con un dispositivo especial para bajar el eje del instrumento a medida que éste va girando, se ha encontrado de excelente reproducibilidad y facilidad de manejo, cuando se usa para determinar la dureza de cristales de guayaba.

2. Los valores obtenidos por el uso de este viscómetro se han comparado con los valores correspondientes obtenidos por el uso del instrumento desarrollado por Tarr y Baker en la Estación Experimental de Delaware.

3. El viscómetro de torsión descrito en este trabajo se recomienda para la determinación de dureza de cristales en laboratorios de control de la industria de conservas y para determinar la dureza de cristales preparados para determinar el grado de las pectinas usadas. Una curva de calibración puede prepararse utilizando una pectina standard y de dicha curva puede determinarse el grado de las pectinas examinadas.

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Correlation at Different Ages Between Nitrogen, Green- and Dry-Matter Contents of Leaves, and Yield of Sugarcane Grown in Sand Culture

J. A. Bonnet, A. R. Riera, and J. Roldán[1]

INTRODUCTION

Sugarcane and nitrogen play important roles in the agricultural economy of the Commonwealth of Puerto Rico. In 1950-51, nearly 45 percent of the total land in crops was devoted to growing sugarcane and 10,501,396 tons of cane were produced with a value of $106,379,000. The total for 1953-54 was 10,879,000 tons with a value of about $104,000,000. About 100,000 tons of nitrogen-bearing materials were imported in 1949-50, at a cost of $4,000,000; this increased to 272,177 tons in 1950-51, and the amount imported was 213,914 tons in 1953-54, costing about $8,000,000.

Nitrogen imports doubled after 1949-50, while sugarcane production remained more or less constant. These figures imply an excessive application of nitrogen to sugarcane above that required for optimum growth. Data from two long-time experiments performed by the Soils Department of the Agricultural Experiment Station of the University of Puerto Rico, one in an acid soil in the humid area and one in a calcareous soil under irrigation in the arid area, have demonstrated that optimum yields of 12 consecutive crops of sugarcane, averaging 45 tons of cane per acre per crop, could be maintained with an annual application of 103 pounds of nitrogen per acre. Most farmers of Puerto Rico generally use two to three times that amount.

REVIEW OF THE LITERATURE

Clements (5, 6)[2] in Hawaii, Bonnet (1), and Capó, *et al.*(4), in Puerto Rico, Innes (8) in Jamaica, and Halais (7) in Mauritius have discussed leaf nitrogen as a suitable index for the control of fertilizer applied to sugarcane. Rouillard (10) proposed the vegetative index, or the weight of 20 sugarcane leaves of the third rank, with the sampling done during the summer months, January to April, at the age of 8 to 10 months for virgins and 5 to 7 months for ratoons.

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[2] Numbers in parentheses refer to Literature Cited p. 108–9.

OBJECTIVES

A suitable index such as is proposed in this paper would be of great assistance to farmers if it enabled them, early in the life of their sugarcane crops, to determine whether additional nitrogen applications would be beneficial. For they could then correct nitrogen deficiencies affecting growth, and this would ultimately lead to large reductions in nitrogen costs.

In this work the low and high levels of nitrogen were known before planting time, because inert sand instead of soil was used as a substratum. Data are presented on correlation studies between the nitrogen content, the green and dry weights of the leaves, and the yield of sugarcane as a means of predicting relative yields when the nitrogen levels in the growth medium varied from deficiency to excess.

EXPERIMENTAL PROCEDURE

Twenty-eight concrete pits, each about 0.00077 of an acre in area, 7 feet 9 inches long, 4 feet 4 inches wide, and from 1 foot 9 inches to 1 foot 11 inches deep, provided with a drainage outlet, were constructed on the floor of a greenhouse with glass and ventilators at the roof, and open at the sides as shown in figure 1. A 3-inch layer of 800 pounds of thoroughly washed, noncalcareous, ½-inch, river gravel was placed on the bottom of each pit; this was covered with 3,000 pounds of acid quartz sand.

The seven randomized treatments used were replicated four times and consisted of variable-unit concentrations of nitrogen, while the other nutrient elements were kept constant at maximum rates. The unit concentration used per pit was 0.03 pound of N, 0.015 pound of P_2O_5, and 0.015 pound of K_2O.

The nitrogen was applied to the sand in the ratio of 90 parts of nitrate to 10 parts of ammonia, as proposed by Trealease and Trealease (11) for a medium around pH 6.0. The ammonia was added as ammonium sulfate, and the nitrate as tetrahydrated calcium nitrate. Seven different levels of nitrogen were studied, each containing 9 units of P_2O_5 and 13 units of K_2O (see table 4, p. 105). The concentrations in parts per million of the other elements were; Ca, 165; S, 124; Mg, 33; Fe, 13; Mn, 1.07; Zn, 0.83; Co, 0.27; B, 0.13.

Sugarcane seeds of the M. 336 variety were disinfected before planting by immersion in hot water at 52°C. for 20 minutes, and were then cut crosswise in one-eye node sections. These sections were planted on September 25, 1952, at a rate of one for each of four holes uniformly spaced in each pit, and were also germinated in a separate sand-bed to supply material for replanting. The plants were irrigated with rainwater when needed, after the drainage had been poured back.

Composite samples of the third, fourth, fifth, and sixth leaves, including

blades and veins, were taken monthly at various growth intervals starting at 3 and ending at 15 months. The green weight of each of these samples was taken, and they were then prepared and analyzed for moisture and nitrogen by a method reported elsewhere (2, 9).

Arrowing started at 13 months. The crop was harvested on December 25, 1953, at the age of 15 months. Weights of cane, sugar, trash, and roots were recorded per treatment and the cane juice was analyzed for juice ex-

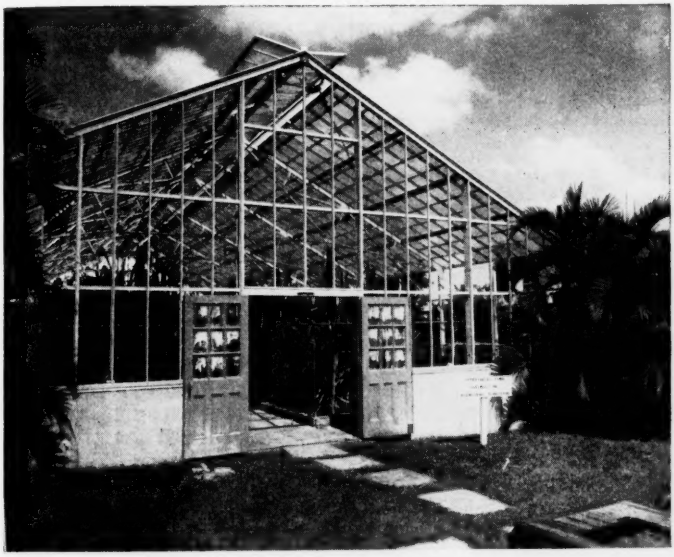

FIG. 1—Sugarcane variety M. 336, 9 months old and just before arrowing, growing in sand culture in the greenhouse used for the experiment.

traction and sugar content. The correlation between the nitrogen content, or the arc-tangent (3) of the nitrogen content of the sugarcane leaves, at the different monthly intervals, and the yield, or relative yield, of the cane were determined for the different nitrogen levels. The relative yields were based on percentages of the maximum yield. Similar correlations were also determined for the green and dry weights of the sugarcane leaves.

RESULTS AND DISCUSSION

The mean nitrogen contents and the green and dry weights of sugarcane leaves at the various monthly intervals are reported in tables 1 to 3. The yields of cane, sugar, trash, and roots, the percentage of extracted sugar

TABLE 1—*Mean nitrogen contents at different crop ages of the leaves of sugarcane variety M. 336 when grown in sand cultures at different nitrogen levels*

Units[1] of N per pit	Percentage of nitrogen (N) in months indicated—												
	3	4	5	6	7	8	9	10	11	12	13	14	15
1.0	2.93	1.33	1.28	1.16	1.09	1.27	1.43	1.43	1.40	1.12	1.30	1.01	1.18
2.5	3.07	1.95	1.74	1.52	1.16	1.22	1.51	1.49	1.37	1.14	1.39	1.19	1.15
4.0	2.96	2.03	1.88	1.54	1.27	1.39	1.45	1.31	1.39	.99	1.11	.95	1.07
5.5	2.92	1.91	1.90	1.82	1.53	1.43	1.71	1.51	1.38	1.11	1.25	1.08	1.03
7.0	3.03	1.99	1.98	1.69	1.70	1.60	1.78	1.61	1.39	1.20	1.32	1.19	1.31
8.5	3.00	2.12	2.05	1.92	1.69	1.75	1.79	1.64	1.53	1.25	1.39	1.29	1.20
10.0	3.10	2.14	2.00	1.91	1.82	1.80	1.92	1.68	1.65	1.35	1.47	1.28	1.29

[1] One unit is equal to 0.03 lb. of N.

TABLE 2.—*Mean green weights at different crop ages of the third, fourth, fifth, and sixth leaves of sugarcane variety M. 336 when grown in sand cultures at different nitrogen levels*

Units[1] of N per pit	Green weight in grams in months indicated—												
	3	4	5	6	7	8	9	10	11	12	13	14	15
1.0	148	178	184	178	213	196	171	194	110	164	120	100	105
2.5	124	171	200	202	250	212	186	211	108	199	116	115	129
4.0	130	174	205	219	235	233	233	227	130	208	146	104	123
5.5	121	184	202	224	270	257	269	236	146	212	135	122	127
7.0	136	190	201	223	288	249	292	281	136	245	151	153	165
8.5	104	155	211	230	312	286	300	292	126	239	174	152	139
10.0	144	172	207	219	315	267	295	284	137	253	199	136	166

[1] One unit is equal to 0.03 lb. of N.

TABLE 3.—*Mean dry weights at different crop ages of the third, fourth, fifth, and sixth leaves of sugarcane variety M. 336 when grown in sand cultures at different nitrogen levels*

Units[1] of N per pit	Dry weight in grams in months indicated—												
	3	4	5	6	7	8	9	10	11	12	13	14	15
1.0	29	44	49	50	62	62	51	48	31	45	34	29	32
2.5	25	41	49	54	73	66	55	52	30	55	32	34	38
4.0	27	42	49	59	68	72	69	59	36	61	43	31	36
5.5	24	45	51	60	74	76	76	60	41	65	39	36	39
7.0	28	47	51	60	78	73	79	70	37	69	43	43	48
8.5	22	39	51	60	82	79	81	73	35	67	49	44	45
10.1	30	41	50	59	85	74	78	70	38	70	55	39	49

[1] One unit is equal to 0.03 lb. of N.

in the cane, and the least significant yield difference between mean treatments are reported for the various nitrogen treatments in table 4. There were significant responses at the 1-percent level to nitrogen in the yields of cane, sugar, trash, and roots.

The percentages of variation explained by correlations between nitrogen, green or dry weights of the third, fourth, fifth, and sixth leaves of the cane, obtained at different ages of the cane, and yields, are reported in table 5, and the coefficients of variability in table 6. The significance of the correlation of the corresponding yield estimate was about the same for all ages between arc-tan nitrogen (3) and relative yield, and between nitrogen per-

TABLE 4.—*Mean cane, sugar, trash, and root yield, and percentage sugar content of sugarcane variety M. 336, with least significant differences between treatment means, when grown in sand cultures in pits at different nitrogen levels*

Treatment No.	Units[1] per pit of —			Mean crop yield in tons per acre of—				Percentage of sugar in cane
	N	P_2O_5	K_2O	Cane	Sugar	Trash	Roots	
1	1.0	9	13	20.0	2.1	5.8	4.0	10.3
2	2.5	9	13	27.8	3.1	8.1	5.9	11.2
3	4.0	9	13	50.1	5.9	11.9	8.4	11.9
4	5.5	9	13	62.8	8.3	13.8	7.5	13.1
5	7.0	9	13	73.8	10.0	16.1	8.1	13.7
6	8.5	9	13	81.3	11.0	19.0	7.7	13.5
7	10.0	9	13	83.1	10.7	19.0	7.3	12.9
L.S.D. between treatment means:								
1-percent point				13.2	1.4	4.0	3.3	1.9
5-percent point				9.6	1.0	2.9	2.4	1.4

[1] One unit equals 0.03 lb. of N and 0.015 lb. of P_2O_5 or K_2O.

centage and yield of cane; so also were the corresponding coefficients of variability. This similarity in the pattern of significance of correlations may be attributed to the low values of leaf-nitrogen percentage (range 0.95 to 3.10, table 1) where the tangent curve is more or less linear.

The significance of correlation and coefficients of variability was about the same between green weight or dry weight and yield of cane, but a higher correlation was obtained by the green weight at 6 months and by the dry weight at 15.

The correlation between leaf-nitrogen percentage and cane yield was not significant at ages of 3, 11, 12, 13, and 15 months; it was significant at the 5-percent level at 10 and 14 months, and at the 1-percent level at 4, 5, 6, 7, 8, and 9 months. The percentage of variation explained for the highest significant correlations between these values varied from 26 to 61, and the

coefficients of variability from 27 to 38. The highest variation explained and the lowest coefficient of variability were obtained when the cane was

TABLE 5.—*Percentage of variation explained by correlation of the yield estimates obtained at different ages of the cane*[1]

Cane age (months)	Leaf arc-tan N and relative yield	Leaf N percentage and cane yield	Green weight and cane yield	Dry weight and cane yield
3	1	3	0	0
4	38**	36**	3	8
5	32**	28**	11	4
6	32**	26**	28**	22*
7	65**	61**	39**	37**
8	52**	50**	56**	28**
9	33**	33**	79**	80**
10	14*	15*	25**	24**
11	8	8	16*	18*
12	8	10	38**	42**
13	0	0	46**	49**
14	23*	22*	14*	16*
15	4	4	11	16*

[1] * Significant at 5 percent; ** significant at 1 percent.

TABLE 6.—*Coefficients of variability of the yield estimates obtained at different ages of the cane*

Cane age (months)	Leaf arc-tan N and relative yield	Leaf N percentage and cane yield	Green weight and cane yield	Dry weight and cane yield
3	44	44	44	44
4	35	35	44	42
5	36	37	42	42
6	37	38	38	39
7	26	27	35	35
8	31	31	30	38
9	36	36	21	20
10	41	41	38	39
11	42	43	41	40
12	42	42	35	34
13	44	44	33	32
14	39	39	41	41
15	43	44	42	41

7 months old. The coefficient of correlation for 7 months was 0.78 for leaf nitrogen and cane yield and 0.81 for arc-tan nitrogen and relative yield, and the regression equations obtained were:

(Yield of cane in tons/acre) = 63.021 (leaf N in percentage) − 35.26

(Relative yield of cane) = 3.567 (arc-tan N in percentage) − 136.32

The correlation between green weight and yield of cane was not significant at the ages of 3, 4, 5, and 15 months; was significant at the 5-percent level at the ages of 11 and 14 months; and significant at the 1-percent level at the ages of 6, 7, 8, 9, 10, 12, and 13 months. Similar results were obtained with dry weight, except that the correlations at 6 and 15 months were significant at the 5-percent level. The percentages of variation explained for the highest significant correlations between green or dry weights and cane yields varied from 28 to 80 percent and the coefficients of variability between 20 and 39 percent. The highest variation explained and the lowest coefficient of variability were obtained when the cane was 9 months old.

The coefficient of correlation for 9 months was 0.89 for either green or dry weight and cane yield. The respective regression equations for green and dry weights were:

(Yield of cane in tons/acre) = 0.394 (green weight of leaf in grams) − 41.24

(Yield of cane in tons/acre) = 1.767 (dry weight of leaf in grams) − 66.53

CONCLUSIONS

The above correlation analyses indicate that the percentage nitrogen content of the third, fourth, fifth, and sixth leaves of sugarcane can be used as an index to predict the yield of cane almost as precisely as the arc-tangent of the nitrogen percentage.

The green and dry weights of these leaves can also be used to predict the yield.

The age of the plant must be taken into consideration for prediction of the yield. Correlations with nitrogen were significant at all ages between 4 to 10 months, and of green or dry weights between 6 to 14 months, for the M. 336 cane variety grown in sand cultures at different nitrogen levels.

SUMMARY

Sugarcane variety M. 336 was grown in sand cultures in 28 pits under cover to study the effect of increasing levels of nitrogen on crop yield and the correlation between the nitrogen, the green- or dry-matter contents, and crop yield. Seven increments of nitrogen, replicated four times, were studied with other nutrients and water at optimum levels.

The yield in tons per acre varied as follows: Cane, 20.0 to 83.1; sugar, 2.1 to 11.0; trash, 5.8 to 19.0; roots, 4.0 to 8.4.

The sugar in the cane varied from 10.3 to 13.7 percent when the crop was harvested at the age of 15 months. There were significant responses to nitrogen at the 1-percent level in the yields of cane, sugar, trash, and roots.

The nitrogen content of the third, fourth, fifth, and sixth leaves varied from 0.95 to 3.10 percent when the cane was between 3 and 15 months old; the green weight of these four leaves varied from 100 to 315 gm. and their dry weight from 22 to 85 gm.

Results indicate that nitrogen, and the green- or dry-matter contents of the cane leaves can be used to predict relative crop yields at specific crop ages. This is illustrated with three formulas derived from the data.

RESUMEN

En el experimento que se informa se sembró caña de azúcar, variedad M. 336, en 28 lisímetros de concreto, bajo techo, que contenían arena como substrato. El objetivo era estudiar el efecto del aumento de siete concentraciones variables de nitrógeno en el rendimiento, y la correlación entre el tonelaje y los contenidos de nitrógeno, materia verde, y materia seca, respectivamente, de las hojas de caña, a diferentes edades. La caña se abonó con cantidades suficientes de fósforo, potasio, y elementos menores; se le aplicó agua periódicamente. Las aguas filtrantes se devolvían a los lisímetros.

Los rendimientos de la cosecha a los 15 meses, en toneladas por acre, variaron como sigue: Caña, 20.0 á 83.1; azúcar, 2.1 á 11.0; paja, 5.8 á 19.0; y raíces, 4.0 á 8.4.

El azúcar en la caña varió de 10.3 á 13.7 por ciento. Hubo respuestas significativas en cuanto al nitrógeno, al nivel del 1 por ciento, en los rendimientos de caña, azúcar, paja, y raíces.

Los contenidos de nitrógeno de las hojas tercera, cuarta, quinta y sexta variaron de 0.95 á 3.10 por ciento entre las edades de 3 y 15 meses; su peso verde varió de 100 á 315 gramos y el peso seco de 22 á 85 gramos.

Los resultados obtenidos indican que los contenidos de nitrógeno, de las hojas analizadas, a las edades entre 4 y 9 meses, se pueden usar como índices para predecir los rendimientos de la caña de azúcar. También pueden usarse como índices de predicción los pesos verdes y secos de las hojas entre los 6 y 13 meses.

Los datos obtenidos con nitrógeno dieron la correlación más alta, a los 7 meses, y los de materia verde o seca, a los 9 meses. Las cuatro fórmulas derivadas, a estas respectivas edades, donde Y expresa el rendimiento de caña en toneladas por acre, fueron las siguiente:

$$Y = 63.021 \text{ (N por ciento en la hoja)} - 35.26$$

$$Y = 0.394 \text{ (peso verde de la hoja en gramos)} - 41.24$$

$$Y = 1.767 \text{ (peso seco de la hoja en gramos)} - 66.53$$

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Practical Evaluation of Garbage Compost as a Fertilizer under Conditions in Puerto Rico

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INTRODUCTION

The possibilities of composting municipal garbage for use as a source of fertilizer elements have been considered lately in Puerto Rico. In 1954, the Economic Development Administration had under study plans of some continental business firms that were interested in establishing garbage-processing plants on the Island with the ultimate purpose of selling the product as a fertilizer. The Soil Conservation Committee of Puerto Rico[2] has shown repeated interest in the possibilities of a method for the disposal of municipal garbage in such a way that additional organic matter and fertilizer elements would be available for farm use. This has been because of the enthusiasm displayed by supporters of the idea that Puerto Rican agriculture is deficient in, and acutely in need of organic manures.

This paper presents data comparing a commercial garbage compost manufactured in continental United States by one of the firms interested in establishing this type of business in Puerto Rico with filter-press cake, a by-product of the sugar mills available to farmers in large quantities during the grinding season. It further compares it with commercial fertilizers as to effect on yields, costs, and bulk.

MATERIALS AND METHODS

The commercial garbage compost and filter-press cake were analyzed for moisture, organic matter, nitrogen, phosphoric acid, and water-soluble potash. Organic matter was determined by loss on ignition and nitrogen by the standard Kjeldahl method. Phosphoric acid was determined by the approved volumetric A.O.A.C. method. Water-soluble potash was determined by means of a flame photometer.

Cost comparisons were made according to prices of fertilizers on August 25, 1955, and the expected selling price of locally produced garbage compost. Calculations of bulkiness were made using sulfate of ammonia, superphosphate, and muriate of potash as standard carriers of the major fertilizer elements.

A Fajardo clay soil was used for the greenhouse evaluation of the com-

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[2] An advisory board to the Soil Conservation Districts, the members of which are appointed by the Governor.

mercial garbage compost in comparison with filter-press cake and inorganic fertilizers. It is an acid, deeply weathered, moderately well-drained nonlatosol of the humid region that occurs in terrace formations. The soil was taken from the upper 6 inches of a site that had been under clean cultivation for several years, but had not been limed. It was air-dried under greenhouse conditions. Its reaction was acid as indicated by a pH of 5.25.

Six kilograms of air-dried soil, screened through a ¼-inch wire net, were placed in each of 57 Mitscherlich's test pots. Nineteen treatments (see table 4, p.7) were set, each replicated three times in a simple randomized-block layout. The treatments included four rates of either commercial garbage compost or filter-press cake: 1, 3, 9, and 27 tons to the acre. Each of these types of treatment was then combined with 100 pounds of each of the major fertilizer elements: Nitrogen, phosphoric acid, and potash. Two additional treatments consisted of 100 and 200 pounds, respectively, of each of the three fertilizer elements. In the control pots the soil was not given any treatment at all.

Thirty-seven pregerminated seeds of Hegari sorghum were planted in each pot. Water was added daily to maintain the soil at approximately field capacity. The pots were randomized in three groups and placed on greenhouse tables keeping the same position until harvest. There was a light attack of the fall armyworm, *Laphygma frugiperda*, when the plants were about 8 inches in height, but it was controlled by spraying with a 1-percent solution of DDT.

RESULTS

The analytical data in table 1 show that the commercial compost used in this experiment was somewhat lower in organic matter than filter-press cake, but both had about the same nitrogen level. The commercial garbage compost was higher in phosphoric acid and considerably higher in potash than filter-press cake.

As shown in table 2, a pound of each of the major fertilizer elements could be obtained reasonably cheap from inorganic sources, but it cost twice as much when supplied from the commercial garbage compost tested in the experiment herein reported, assuming a selling price of $30 per ton of compost.

Table 3 gives an idea of the relative bulkiness of a commercial garbage compost with a given analysis as compared with a fertilizer having an identical composition as the compost. Ten 200-pound bags are needed to pack a ton of this compost while the same fertilizer value can be packed in only 3¼ bags of inorganic fertilizers.

Table 4 gives experimental data obtained from the pot test conducted under greenhouse conditions using Hegari sorghum as an indicator plant.

The data disclose that additions of commercial compost or filter-press cake increased the dry-matter yields of Hegari sorghum from less than 14 gm.

TABLE 1.—*Comparative chemical data on certain constituents of a commercial garbage compost and filter-press cake*[1]

Material	Organic matter	Nitrogen	Phosphoric acid	Water-soluble potash
Commercial garbage compost	36.42	2.28	2.87	4.71
Filter-press cake	39.46	2.27	2.24	.44

[1] All determinations expressed as percentage of dry weight. The compost contained 10.04 and the filter-press cake 12.34 percent of moisture.

TABLE 2.—*Cost analysis in cents per pound of essential major nutrients in a commercial garbage compost and in inorganic fertilizers*

Nutrient	Cost in indicated source	
	Commercial garbage compost[1]	Inorganic fertilizers[2]
Nitrogen	26	13
Phosphoric acid	18	9
Water-soluble potash	8	4

[1] Assuming a selling price of $30 a ton. In Oakland, Calif., the price was $34 a ton, in Chicago it was $50 or $60 a ton.

[2] According to prices quoted by a local fertilizer concern, Aug. 25, 1955.

TABLE 3.—*Quantity of ordinary commercial fertilizer necessary to provide the nutrient equivalent in a ton of commercial garbage compost*

Nutrient	Compost analysis	Quantity of nutrient in a ton of compost	Fertilizer	Purity	Equivalent quantity needed
	Percent	*Pounds*		*Percent*	*Pounds*
Nitrogen	2.28	45.6	$(NH_4)_2SO_4$	21	217
Phosphoric acid	2.87	57.4	Superphosphate	20	287
Water-soluble potash	4.71	94.2	KCl	60.5	155

per pot in the controls to approximately 32 and 24 gm. per pot, respectively, when inorganic fertilizer elements were omitted. However, when the soil was dressed with nitrogen, phosphoric acid, and potash at the rate of 100 pounds to the acre, the application of either compost or filter-press cake did not increase yields significantly. A total yield of about 39 gm. per

pot was obtained when the major fertilizer elements only were applied at a rate equivalent to 100 pounds to the acre of each. The application of the highest amount (27 tons to the acre) of compost and filter-press cake, in conjunction with the fertilizer elements at the rate previously indicated,

TABLE 4.—*Dry-matter yields of Hegari sorghum in a greenhouse test comparing the effect of commercial garbage compost, filter-press cake, and inorganic fertilizers at different rates of application*

Per acre applications of—			Garbage commercial compost used per acre	Filter-press cake used per acre	Mean dry-matter yields per pot
Nitrogen	Phosphoric acid	Potash			
Lb.	*Lb.*	*Lb.*	*T.*	*T.*	*Gm.*
0	0	0	0	0	13.66
0	0	0	1	0	15.33
0	0	0	3	0	20.00
0	0	0	9	0	23.00
0	0	0	27	0	32.30
100	100	100	0	0	39.30
100	100	100	1	0	35.00
100	100	100	3	0	34.00
100	100	100	9	0	45.67
100	100	100	27	0	43.67
0	0	0	0	1	16.67
0	0	0	0	3	13.67
0	0	0	0	9	22.67
0	0	0	0	27	23.67
100	100	100	0	1	32.00
100	100	100	0	3	36.00
100	100	100	0	9	34.67
100	100	100	0	27	42.00
200	200	200	0	0	40.67
L.S.D at the 5-percent level					4.74
L.S.D. at the 1-percent level					6.36

TABLE 5.—*Analysis of variance of the yield data obtained in a greenhouse experiment comparing the effect of commercial garbage compost, filter-press cake, and inorganic fertilizers*

Source	Degrees of freedom	Sum of squares	Mean square	F
Total	56	6,492.32		
Replications	2	70.85		
Treatments	18	6,121.32	340.35	45.51[1]
Error	36	295.15	8.20	

[1] Highly significant.

produced nonsignificant increases of only 4.37 and 2.70 gm. per pot, respectively.

Data on the sum of the squared deviations of the dry-matter yields obtained in this experiment are reported in table 5.

DISCUSSION

In Puerto Rico filter-press cake can be obtained free from the sugar mills, the only expenses to the farmer being those involved in transportation and field application. The value of the extra phosphoric acid and potash that the commercial garbage compost tested has over the filter-press cake can be calculated at only $5 per ton. It has been estimated that, under present conditions, a ton of compost from garbage locally produced would have to sell at a price of about $30 for the plant to function at a reasonable economic level. However, it cannot be expected that farmers will be willing to pay such prices for compost while they can obtain the same fertilizer equivalent at much reduced rates.

One of the industrial firms that was seriously considering establishing this business in the Island required that the Commonwealth Government guarantee an annual sale of 10,000 tons of compost, thus tying the Government up with an investment of some $300,000 annually. The total annual output of the proposed plan was to be about 16,000 tons. At the proposed price this product would have hardly any outlet except in some special cases, such as for small gardens, pots, greenhouses, and among those with the hobby of growing ornamentals. The product might also find a limited outlet in nurseries with high-priced stock, for flowers, and on lawns. There are absolutely no possibilities of marketing 16,000 tons yearly at the indicated price levels. In an emergency such as wartime, such a plant might do some business, if the shipments of fertilizers were adversely affected, but even then a price of $30 a ton would be too high.

In a soil like the Fajardo clay used in the pot tests reported in table 2 neither filter-press cake nor garbage compost seemed to be a paying proposition as compared to commercial fertilizer elements, at least to judge from the yield differences. The fertilizer elements applied per acre would cost some $26 and the yields obtained could not be increased practically by using the organic materials tested. The use of 27 tons to the acre of garbage compost would not even result in yields as high as those obtained with 100 pounds of each of the major fertilizing elements, while the cost of the material would be some $810, not including the considerable expenses that the transportation and application of such bulky material would entail.

There would be no present justification for the establishment of plants to dispose of town wastes if their success depended on the eventual possi-

bility of selling the product at prices even 50 percent below the estimated selling price of $30 per ton.

Insofar as soil organic-matter levels are concerned it might be well to stress further that many of the soils of Puerto Rico are well or moderately well supplied with organic matter. Smith, *et al.* (9)[3], have shown that the organic-matter contents of these soils are at least as high as those of many of the best soils of the Temperate Zone. Furthermore, Lugo-López, *et al.* (4), have demonstrated experimentally that additions of low-nitrogen organic-matter sources to normal latosols with an open, porous structure are of rather doubtful value. Yields of sugarcane and pineapple were not influenced by such soil treatments (2). On the other hand, the same authors (4) reported that low-nitrogen organic-matter additions to heavy-textured nonlatosols with an unfavorable physical condition resulted in increased organic-matter levels, and that sugarcane and pineapple yield increases could be attributed to these treatments (2).

A better utilization of farm resources can be of great help in organic-matter maintenance and soil build-up on many Puerto Rican farms. Sugarcane farmers, for instance, have a steady potential supply of organic matter in sugarcane trash (5). Coffee farms, with the large amount of litter from the coffee and the shade trees, in addition to the pulp from coffee berries, have hardly any further need of additional organic-matter sources. Grasslands and forests should present no problems. The use of grasses and legumes in suitable rotations, as well as the use of green manures and cover crops will be of great assistance in other areas where large amounts of organic residues are not normally available, if and when they are needed.

In those cases where additional organic-matter sources might be needed, farmers could supply these at a relatively low cost through the use of filter-press cake (3), bagasse, animal manures, green manures (1), and crop residues. The preparation of composts on the farm might be of practical value in these cases. In this connection, it might be well to mention that the addition of bacteria of the types isolated by Pfeiffer (6, 7) may considerably speed up the humification of farm compost piles. Through the use of these bacteria the process of composting can be completed in a very short time. In the same fashion, these bacteria may be of value in speeding up the decomposition of cover crops and crop stubble. This might save time in starting new crops.

There is no question as to the utility of garbage-processing plants or techniques for the sole purpose of disposal of town wastes. Use of such a technique, using bacteria as a starter to speed up decomposition, was recently initiated at Yabucoa, in southeast Puerto Rico, through the efforts of the

[3] Numbers in parentheses refer to Literature Cited p. 117.

Department of Health[4]. There is a rather large plant successfully operating at Wyster in the Netherlands. There are probably others in Europe. Reports are that the Dutch concern functions on an economically sound basis and is probably justified because Dutch agriculture is claimed to be short of organic manures (8). In the United States there are plants in operation at Long Island, N. Y., Oakland, Calif., Chicago, Ill., and probably at other locations. The price quoted by the Oakland plant is $34 per ton and the Chicago plant sells at $50 to $60 a ton. On farm scale, this is obviously an uneconomical business.

Under Puerto Rican conditions there is hardly any room for such plants other than for garbage-disposal purposes.

SUMMARY

Data are presented to show the relative economic inefficiency of garbage compost when compared with inorganic fertilizers and filter-press cake. The content of potash and phosphoric acid is higher in the garbage compost than in the filter-press cake, but the former is slightly lower in organic matter. They both have about the same nitrogen level.

In a greenhouse experiment it was found that additions of garbage compost and filter-press cake increased sorghum yields in the absence of inorganic fertilizers. However, when nitrogen, phosphoric acid, and potash were applied at the rate of 100 pounds to the acre each the application of either compost or filter-press cake did not increase yields significantly.

Under the conditions that prevail in Puerto Rico there is hardly any need for plants to process town wastes for fertilizer production. At current price levels, the cost of commercial garbage compost would be so high in relation to its fertilizer-element value, that it could not compete with commercial inorganic fertilizers. The product is so bulky that hauling and application costs would be too high under normal conditions. Furthermore, the organic-matter levels of Puerto Rican soils are not so low as to justify the use of such commercial composts. Under average farm conditions there are more practical means of raising soil organic-matter levels, if and when necessary.

RESUMEN

Se presentan datos que comparan el efecto de las aplicaciones de un composte comercial hecho de basuras con el de la cachaza y el de los abonos inorgánicos.

El contenido de potasa y ácido fosfórico del composte es más alto que el

[4] Experimentan método para eliminar basuras, *El Mundo*, San Juan, P. R. Sept. 1, 1955.

de la cachaza, pero el composte tiene un poco menos materia orgánica. Ambos tienen más o menos el mismo contenido de nitrógeno.

En un experimento hecho en el invernadero se encontró que aplicaciones de composte o cachaza aumentan los rendimientos de sorgo en la ausencia de abonos inorgánicos. Sin embargo, cuando se aplicó nitrógeno, ácido fosfórico y potasa, a razón de 100 libras por acre en cada caso, la aplicación de composte y cachaza no aumentó los rendimientos significativamente.

Bajo las condiciones en que se desenvuelve la agricultura en Puerto Rico, no es aconsejable ni hay necesidad por ahora de establecer factorías para convertir las basuras en material fertilizante. En contra del establecimiento de tales empresas, se puede argumentar que, unidad por unidad de nutrimentos, el costo de los obtenidos de estos compostes comerciales resultarían tan altos que no podría competir con el de los abonos inorgánicos. Además, el producto sería tan voluminoso que los costos de acarreo y aplicación resultarían muy altos. Las deficiencias de materia orgánica en la mayoría de los suelos de Puerto Rico no son tan agudas como para justificar el uso de productos de esa naturaleza. Se indica que hay medios más prácticos para mejorar esos niveles, si fuere necesario.

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Bacterial Flora of Foot Rot: Report of Findings in 100 Cattle

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INTRODUCTION

Foot rot of cattle is a disease of economic importance occurring endemically in Puerto Rico, especially in the northern part of the Island. The disease is prevalent during rainy periods when the mud macerates the feet of cattle, predisposing them to infection. In Puerto Rico, animals of the Holstein breed, extensively used for milk production, are especially susceptible to this condition.

Foot rot is an inflammation of the tissues of the feet which may lead to pus tracts and death of foot tissue, producing lameness, which may become permanent if not attended promptly. When thus affected, the animal loses appetite, and milk production and breeding efficiency are greatly lowered.

REVIEW OF THE LITERATURE

In spite of the economic importance of this disease a bacteriological study of foot rot lesions is lacking. Numerous references in the literature imply that *Spherophorus necrophorus* is the causative agent (1,2,3,4,5,6) [2] but definite proof of this is lacking. The evidence lies mostly in the study of direct smears from scrapings of foot rot lesions (7). This is not conclusive since only a small percentage of the smears examined (7) are reported as showing organisms with the morphology of *S. necrophorus* (*Actinomyces necrophorus*).

Since the etiology of the disease is rather obscure, treatment has been undertaken on an empirical basis. There are various reports on the use of sulfa drugs (8,9,10,11,12,13,14,15,16) some being favorable, while other workers report that local treatment is as good as any (17). Emphasis has also been placed on preventive measures (1,18). However, since preventive methods are not always feasible or practical, a study of the bacteria found in foot rot lesions should give a more complete picture of the organisms involved, and therefore indicate the drugs or antibiotics likely to be most useful in treating this condition.

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[2] Numbers in parentheses refer to Literature Cited, p. 124.

MATERIALS AND METHODS

SPECIMENS

Specimens were taken from diseased cattle at 10 cattle farms located in Hato Rey, Bayamón, Dorado, Carolina, and Canóvanas. The procedure was as follows: The diseased foot was immobilized and washed thoroughly with tapwater to remove as much dirt as possible. The interdigital space was further cleaned with cotton and 70-percent alcohol. A sterile cotton swab was then introduced into the wound or, when the lesion was small, it was just rubbed over the lesion site, a smear was made on a sterile slide, and the specimen was then immersed in a tube of thioglycollate broth enriched with 10-percent beef serum. The smear was stained by Gram's method and examined under the microscope.

Several types of lesions were encountered. The great majority involved swelling of the tissues between the claws, associated with an open wound or scratch.

CULTURE

The thioglycollate-broth culture was incubated at 37°C. and as soon as growth was observed, smears were made and stained by Gram's method. If no growth was observed the inoculated broth culture was further incubated for 7 days, after which it was discarded if still sterile.

The thioglycollate-broth culture was streaked on duplicate plates of rabbit-blood agar. One plate was incubated aerobically while the other was placed under anaerobic conditions in an atmosphere of nitrogen. To insure growth of all anaerobic organisms an additional plate of Brewer anaerobic blood agar was also streaked and incubated. After a 24- to 48-hour incubation, colonies from the various plates were examined and described.

Isolated colonies were picked to tubes of heart-infusion broth, or trypticase-soy broth in the case of aerobic organisms, and to the thioglycollate medium when the colonies grew under anaerobic conditions. Purity plates were performed on rabbit-blood agar or Brewer anaerobic-blood agar, and when a pure culture was obtained, fermentation and other tests were carried out for classification. Such organisms as *Bacillus alkaligenes*, *Proteus vulgaris*, and *B. coli* were encountered irregularly as contaminants and discarded.

PATHOGENICITY TESTS

Pathogenicity tests were carried out on several cultures of pathogenic or potentially pathogenic species of organisms. White mice were injected with 0.1 to 1 cc. of culture by the intraperitoneal or subcutaneous route. A few tests were also carried out on guinea pigs.

SENSITIVITY TESTS

Sensitivity tests were carried out on a number of representative cultures of the more important organisms isolated. An 18- to 24-hour broth culture of the organism was evenly spread on the surface of a blood-agar plate and Bacto sensitivity disks containing aureomycin, bacitracin, chloromycetin, penicillin, dihydro-streptomycin, polymyxin B, terramycin, erythromycin, and magnamycin were placed on the surface with sterile precautions about 4 cm. apart. The plates were incubated aerobically or anaerobically, depending on the organism tested, until growth developed. They were then examined for zones of inhibition of growth.

TABLE 1.—*Bacteria isolated from 108 lesions of foot rot in 58 recent and 50 old cases*

Organism	Recent cases (58)	Old cases (50)	Percentage of total number of specimens (108)
S. necrophorus	7	0	6.4
C. pyogenes	7	4	10
Clostridium spp.[1]	18	11	26.8
Streptococci	33	26	54
Micrococci	9	10	17
B. cereus	33	23	52
Bacteroides melaninogenicus	8	3	10
Ps. aeruginosa	3	0	3

[1] *Cl. sporogenes* and *Cl. bifermentans*, mostly.

RESULTS

A total of 108 specimens was obtained from 100 head of cattle on 10 cattle farms located at Hato Rey, Bayamón, Dorado, Carolina, and Canóvanas. Fifty-eight of the specimens were from recent cases 1 day to 2 weeks in duration, while 50 specimens were taken from cases of more than 2 weeks duration. The results of the cultural studies are shown in table 1. Cases of more than 2 weeks duration are recorded in the table as "old" cases. In this table the number of positive cultures for the species listed is shown in the first two columns under "recent" and "old" cases. The right-hand column gives the percentage of positive cultures for the total number of specimens taken.

It will be seen that *S. necrophorus*, the organism commonly suspected of being the cause of cattle foot rot, was found in only 6.4 percent of the total number of specimens taken. *B. cereus* and Streptococci appeared most frequently of all species in the table. *B. cereus*, however is very abundant in soil and almost all the Streptococci found were nonhemolytic. A mixed bacterial flora was cultured from all cases.

An attempt was made to correlate the presence of Gram-negative bacilli showing the characteristic morphology of *S. necrophorus* in direct smears from foot rot lesions with the number of cultures positive for *S. necrophorous*. It was observed in 11 specimens showing suspicious bacilli in the direct smears that 5 gave a culture positive for *S. necrophorous*. On the other hand, in 2 specimens from which a positive culture was obtained, the smear from the lesion did not disclose any organisms morphologically resembling *S. necrophorus*.

Pathogenicity tests were carried out on a number of strains of *S. necrophorus*, *C. pyogenes*, *Ps. aeruginosa*, and *B. cereus*. The results of these tests are shown in table 2. Although *B. cereus* is one of the more frequent

TABLE 2.—*Pathogenicity of 4 bacterial species isolated from cases of foot rot*

Species tested[1]	Number of strains tested	Number found pathogenic
S. necrophorus	4	1
C. pyogenes	4	4
Ps. aeruginosa	2	2
B. cereus[2]	7	1

[1] Pathogenicity of *S. necrophorus*, *C. pyogenes*, and *B. cereus* was tested on white mice; *Ps. aeruginosa* was tested on guinea pigs.

[2] Pathogenicity tests were carried out on several strains of *B. cereus* since this species occasionally has been reported as pathogenic (19).

inhabitants of soil and dirt and is commonly regarded as a contaminant, seven strains were tested for pathogenicity since there have been reports (19) of pathogenic effects produced by this organism. However, only one of the strains proved actually pathogenic. In each case the pathology was characteristic for the species tested. All strains of *C. pyogenes* also produced a hemolysin active on rabbit red cells.

A limited number of sensitivity tests were carried out by the paper-disk method using nine different antibiotics and several strains of *C. pyogenes*, *Cl. bifermentans*, *Cl. sporogenes*, *S. necrophorus*, *Ps. aeruginosa*, and one pathogenic *B. cereus* strain.

The majority of strains of *C. pyogenes* tested were completely susceptible *in vitro* to aureomycin, bacitracin, terramycin, erythromycin, and magnamycin. All strains of the Clostridia (*Cl. bifermentans* or *sporogenes*) were susceptible to aureomycin, chloromycetin, and terramycin. Most strains of *S. necrophorus* were susceptible to aureomycin and chloromycetin. Results with other antibiotics were not as definite. As expected, the wide-spectrum antibiotics were most effective against the several species of bacteria tested.

DISCUSSION

A study of the bacterial flora obtained from cases of cattle foot rot has been lacking to the present. The study here reported was undertaken with the purpose of ascertaining whether a single bacterial species was involved in at least the majority of cases, or whether predominance of a few species occurred in most of the cases studied. A mixed bacterial flora was found in all lesions cultured, whether recent—1 day to 2 weeks' old—or old. Thus no particular species or group of organisms predominated in any case.

The cultivation of material from the diseased feet of cattle is attended by the presence of numerous contaminants, since the feet are continuously exposed to feces, urine, soil, and dirt. Organisms present in these sources have easy access to the interdigital space and to any abrasions or bruises in the feet. In spite of precautions taken in having the feet as clean as possible, and in taking the specimens only from the lesion site, many of the organisms appearing were common contaminants and were discarded as such. Attention was therefore directed to the isolation and cultivation of organisms belonging to genera which most probably would contain the offender or offenders: Spherophorus, Clostridium, Bacteroides, Streptococcus, Staphylococcus, Corynebacterium, and Pseudomonas. Many organisms belonging to these genera cause a variety of diseases in cattle and may be found in materials such as feces, dirt, manure, and urine which come in direct contact with wounds or abrasions on the feet of cattle.

Perhaps the most interesting finding was that *S. necrophorus* was actually isolated from the diseased feet of cattle but only from fresh lesions. It was found in only 6.4 percent of all specimens. It is possible that this organism may have been present in a larger number of specimens and may have been missed in some because its cultivation is difficult, the more so because of overgrowth on the part of the numerous contaminants encountered.

Another interesting finding was the isolation of *C. pyogenes* from 10 percent of all specimens from both recent and old lesions. To our knowledge this is the first time *C. pyogenes* has been connected with foot rot in any way, and it may well play a role in at least some cases.

As mentioned before, *S. necrophorus* has been mainly suspected to be the most common offender in cattle foot rot. The finding of long Gram-negative filaments and pleomorphic bacilli, or short Gram-negative bacilli showing bipolar staining in smears, has been regarded by some as sufficient evidence for incriminating *S. necrophorus*. Actually quite a few species of Gram-negative bacilli, such as *Proteus vulgaris* and *B. coli* occasionally show these long filamentous forms in direct smears from lesions (as stated in a personal observation by J E. Pérez). Therefore the only way of actu-

ally proving the causative agent to be *S. necrophorus* would be by its occurrence in pure culture, or, at least as a predominant species in mixed culture. In our study the suspicious Gram-negative filaments were found in 11 specimens, but only 5 of these actually yielded *S. necrophorus* and then only in mixed culture.

Bacteroides melaninogenicus was found in 11 percent of all lesions. Dack (20) mentioned this organism as one of the species associated with ulcerative processes in human beings.

The limited number of sensitivity tests carried out *in vitro* and the results from the cultural studies suggest that wide-spectrum antibiotics should be employed in the management of foot rot, to insure proper coverage of most of the organisms involved.

From the results obtained it is evident that a low incidence of pathogens was encountered. It may be that organisms usually regarded as nonpathogens *per se* may exert a pathogenic effect when found associated in certain combinations, but this is a matter of speculation which would have yet to be proven in the particular condition of cattle foot rot.

SUMMARY

One hundred and eight specimens were obtained for bacteriological study from 100 head of cattle affected with foot rot on 10 cattle farms located in Hato Rey, Bayamón, Dorado, Carolina, and Canóvanas. Smears were made on sterile slides and stained by Gram's method. Aerobic and anaerobic cultures were also made with the specimens.

Gram-negative bacilli morphologically resembling *S. necrophorus* were encountered in about 10 percent of the smears, yet the organism itself was cultured from only half of the specimens having positive smears. *Bacillus cereus*, Streptococci, and Clostridia were found in many specimens. *Corynebacterium pyogenes* and *Bacteroides melaninogenicus* each had an incidence of 10 percent of the total number of specimens. *Pseudomonas aeruginosa* was cultured from only 3 percent of all specimens.

It was concluded from limited *in vitro* tests that wide-spectrum antibiotics were most active against these organisms. However, *Ps. aeruginosa* was resistant to all the antibiotics tested.

RESUMEN

Se tomaron 108 muestras de ganado afectado por la podredumbre de la pezuña (mazamorra) para estudios bacteriológicos. Se hicieron frotis, los cuales se tiñeron al Gram. Con las muestras tomadas se hicieron también cultivos para aerobios y anaerobios. Bacilos Gram negativos, morfológicamente similares al *Spherophorus necrophorus*, fueron notados como en un 10 por ciento de los frotis. El *Spherophorus necrophorus*, sin embargo, pudo cultivarse sólamente de la mitad de las muestras que dieron frotis positivos.

En muchas muestras se encontraron organismos de los géneros Streptococcus, Clostridium y el *Bacillus cereus*. Tanto el *Corynebacterium pyogenes* como el *Bacteroides melaninogenicus* tuvieron individualmente la incidencia de un 10 por ciento del número total de muestras. El *Pseudomonas aeruginosa* se encontró sólo en un 3 por ciento de las muestras. Un número limitado de pruebas de sensibilidad mostró que la mayoría de estos organismos fueron susceptibles a los antibióticos de gran radio de acción, como la terramicina, cloromicetina y aureomicina. El *Ps. aeruginosa* fué resistente a todos los antibióticos probados.

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RESEARCH NOTES

EVIDENCE OF THE PRESENCE OF SUGARCANE-MOSAIC VIRUS IN THE ROOTS OF INFECTED SUGARCANE PLANTS

In the course of preliminary experiments designed for the purpose of culturing, *in vitro*, roots of sugarcane infected with sugarcane mosaic it was necessary to determine whether the virus was definitely present in the roots of infected canes. Interestingly enough we have not been able to find in the available literature any information or report as to the actual determination of the sugarcane virus in the root tissue. In order to clarify this point an experiment was carried on in which pieces of surface-sterilized sugarcane cuttings from mosaic-diseased plants were placed inside moist glass chambers to root. When the roots were about 2 to 3 inches long they were carefully cut with scissors and macerated in a mortar with a little sterile water. The extract was inoculated with the aid of carborundum on sorghum (*Holcus sorghum* l.)[1] seedlings. Of a total of 100 plants inoculated 6 became infected with mosaic, thus confirming the presence of the virus in the roots.

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[1] Adsuar, J., On the physical properties of sugarcane-mosaic virus, *Phytopath.* **40** (2) 214–6, 1950.

TRANSMISSION OF CUCUMBER MOSAIC (CUCUMBER VIRUS 9) FOUND IN PUERTO RICO TO CARICA PAPAYA

In February 1953, during the course of some experiments carried on in relation to the cucumber mosaic (cucumber virus 9) found in the Island,[1] plants of *Carica papaya*, *Cucumis sativus* var. Black Diamond, and *Phytolacca decandra* were inoculated with juice from a tobacco plant infected with cucumber virus 9. About 2 weeks later one of the inoculated papayas displayed symptoms of vein-clearing and mottling. The inoculated cucumbers showed symptoms characteristic of virus 9 infection and *P. decandra* necrotic spots on the inoculated leaves. Several weeks later the affected papaya began to show symptoms indistinguishable from those produced in that host by the papaya-mosaic virus.[2] The possibility of establishing the relationship between the viruses attacking two such unrelated hosts

[1] Adsuar, J. and Cruz Miret, A., Virus diseases of cucumbers in Puerto Rico, Tech. Paper No. 6, Agricultural Experiment Station, University of Puerto Rico, 1950.

[2] Adsuar, J., Studies on virus diseases of papaya (*Carica papaya*) in Puerto Rico, Tech. Paper No. 1, Agricultural Experiment Station, University of Puerto Rico, 1946.

was considered of interest. However, other problems interfered with the study and it was accordingly postponed.

This year we have successfully accomplished the transmission of cucumber virus 9 to papaya. Cucumber seedlings previously inoculated with cucumber virus 9, and showing unmistakable symptoms of mosaic, were macerated, and the extracted juice inoculated into healthy papaya and cucumber plants by rubbing with carborundum. Of various transmission experiments performed only one succeeded. Three out of nine papayas inoculated showed symptoms characteristic of papaya mosaic. All the cucumbers inoculated became diseased. The infected papayas began to manifest preliminary symptoms of vein-clearing and mottling in about 3 weeks, and the characteristic leaf distortion and malformation were noticeable about a month later. So far we have been unable to retransmit the virus from papaya to cucumber. Our experience seems to indicate that the cucumber virus 9 is very difficult to transmit from cucumber to papaya by rubbing the crude juice in with carborundum.

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A VIRUSLIKE DISEASE OF SORGHUM IN PUERTO RICO

In July 1954, two stools of Merker grass, *Pennisetum purpureum* var. Merkeri collected at the Lajas substation and presenting symptoms resembling a viruslike disease were brought to the attention of the writer by Efraín Boneta of the staff of this Station.

The most conspicuous symptom of the disease was a creamy-yellow freckling and striping of the leaves, especially the top ones. The stripings began first to appear at the base of the still unfurled topmost leaf, and extended upward as parallel contiguous yellowish streaks. Reddish specks apparently due to the necrotization of the chlorotic tissue were to be seen in most cases, especially in the region close to the midrib. In severe cases many of the top leaves did not unfurl properly but remained close and twisted. The whole plant was badly stunted and the shortened internodes imparted a bunchy-top aspect to it.

Cultures made from affected tissues yielded only a yellowish, unidentified bacterium and two fungi belonging to the genus *Curvularia* and *Colletotrichum*. So far none of the isolated organisms has proved to be pathogenic when inoculated on healthy Merker plants. Attempts to transmit the disease to healthy Merker, sorghum, and sugarcane plants by juice inoculation with the aid of carborundum, or by pin-pricking, using infectious juice, have so far been unsuccessful.

The possibility that the disease might be due to 2-4D or other herbicide injury was discarded when an inquiry made at the time failed to disclose

any applications either in the plantation or its vicinity. Also it is difficult to reconcile with the above-mentioned view the fact that only two plants among many were affected. Nutritional disturbances were also ruled out when the disease condition persisted after transplanting the stools to a rich, well-manured soil.

The disease condition is transmitted serially through cuttings and is accompanied by a progressive decline in the vigor of the stools. Immersion of the affected cuttings in water at 52°C. for 20 minutes resulted in the production of healthy plants.

We have been unable to find in the available literature reports of a disease of *P. purpureum* var. Merkeri which corresponds to the one under study. However, Cheriam and Kylasam[1] described two diseases, "freckled yellow" and "stripe" of sorghum in India, which have many points of similarity with the disease reported in this paper.

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[1] Cherian, M. C., and Kylasam, M. S., Preliminary studies on the "freckled yellow" and "stripe" diseases of Cholam, *Proc. Assn. Econ. Biol., Coimbatore*, 1936, **4** 57–63, 1937.

A REPORT ON TWO SPECIES OF TOMATO RESISTANT TO THE TOMATO DISEASE KNOWN AS "TISIS" IN PUERTO RICO

In a previous note[1] the author reported the presence in Puerto Rico of a virus disease of tomatoes known locally as "tisis". The disease markedly affects the development and fruit production of the plant and, although its incidence has been very low up to now, it might constitute a serious menace to tomato-growing in the Island if it ever attained epidemic proportions. The disease is transmitted only by grafting and, symptomatologically, it closely resembles the Brazilian curly top of tomatoes as described by Bennett and Costa.

The curly top virus (*Ruga verrucosans* Carsner and Bennett) causes very severe losses of tomatoes in some sections of the United States. Other strains of the virus occur in some parts of South America. Unfortunately no marked resistance has yet been incorporated in a variety of good commercial quality.

Doolittle[2] has recently reviewed the literature concerning the use of resistant varieties and species of tomato in the production of crosses with high resistance to curly top. He cited the varieties Red Peach and Ojo de Venado,

[1] Adsuar, J., A disease of tomato in Puerto Rico resembling the Brazilian top of tomatoes, *J. Agr. Univ. P. R.* **39** (2) 113–4, 1955.

[2] Doolittle, S. P., The use of wild Lycopersicon species for tomato disease control, *Phytopath.* **44** (8) 409–15, 1954.

both lacking a very high level of resistance, and the species *L. peruvianum* var. *humifusum*, *L. peruvianum* var. *dentatum*, and *L. chilense* as possessing a high resistance to curly top.

In view of the similarity already noticed between our tomato "tisis" disease and the strains of curly top affecting tomatoes in South America we decided to test some of the curly top-resistant species of tomatoes reported in the literature against our tomato virus.

Seeds of *Lycopersicum peruvianum* var. *dentatum* and *L. glandulosum*, two species reported as resistant to curly top, were kindly supplied by M. M. Hoover of the USDA Regional Plant Introduction Station, Ames, Iowa. Also included in the tests, although not mentioned in the literature as resistant, were the species *L. hirsutum* and *L. glabratum* obtained through the courtesy of A. Sawant of the Plant Breeding Department of this Station.

All the species tested, with the exception of *L. hirsutum* and *L. glabratum* that were obtained from rooted cuttings, were grown from seed in the greenhouse in 6-inch pots. The plants were side-grafted when about 1½ feet tall with scions taken from *L. esculentum* var. Marglobe tomatoes showing symptoms of "tisis". Reciprocal grafts were also made using diseased Marglobe plants as stock. The plants were kept in the greenhouse for at least 1 month after grafting.

Both *L. hirsutum* and *L. glabratum* proved fully susceptible to the virus. No trouble was experienced in grafting those two species on infected Marglobe tomatoes. On the other hand, the majority of the grafts performed with *L. peruvianum* var. *dentatum* and *L. glandulosum* did not take. However, no symptoms of the disease developed in those scions that were grafted and then grew successfully. It is concluded that both *L. peruvianum* var. *dentatum* and *L. glandulosum* apparently are not affected by the virus causing the "tisis" of tomatoes in Puerto Rico.

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(Continued from inside front cover)

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